PATENT

**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**
**(Case No. 03-284-E)**

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| In the Application of: | ) | |
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| Costa et al. | ) | Before the Examiner: Halvorson, M. |
| Serial No. 10/809,144 | ) | |
| | ) | Group Art Unit: 1642 |
| Filing Date: March 25, 2004 | ) | |
| | ) | |
| For: Methods of Inhibiting Tumor Cell Proliferation | ) | Confirmation No.: 7397 |
| | ) | |

Mail Stop AMENDMENT
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

**DECLARATION OF CO-INVENTOR I-CHING WANG UNDER 37 C.F.R. §1.132**

I, I-Ching Wang, declare as follows:

1. I am a citizen of Taiwan, residing at 727 W. Martin Luther King Drive, Apt. 1005W, Cincinnati, OH 45220.

2. I am currently a Research Associate, Division of Neonatology, Cincinnati Children's Hospital Medical Center. My *Curriculum vitae* was submitted to the Patent Office as Exhibit A to the Declaration under 37 C.F.R. 1.132 filed October 26, 2007 ("the October 26 Declaration").

3. I am the co-inventor of the instant U.S. Patent Application Serial No. 10/809,144, filed March 25, 2004.

4. I am knowledgeable in tumor biology in general, and particularly in the molecular biology of FoxM1B and p19ARF proteins.

5. I hereby declare that FoxM1B is expressed in most proliferating cells, particularly proliferating tumors cells, from different origins. Further, because of p19ARF's inhibitory effects on FoxM1B activities, the p19ARF 26-44 peptide would have broad therapeutic benefits against different types of tumors.

6. It was known at the time of the invention that FoxM1B is commonly expressed in a variety of embryonic and adult proliferating cells. The instant application describes that FoxM1B is expressed in embryonic liver, intestine, lung and renal pelvis, and adult proliferating cells of the thymus, testis, small intestine and colon. See page 3, lines 13-18 of the instant specification. Ye et al. has shown that FoxM1B is expressed in proliferating epithelial as well as mesenchymal cells. See Mol. Cell. Biol. 17:1626-1641 (1997) (Exhibit A). Although FoxM1B is not expressed in postmitotic differentiated cells, its expression is reactivated in the liver during liver regeneration. *Id.*

7. Additionally, FoxM1B is expressed in numerous actively-proliferating tumor cell lines of different origins. See the disclosure in the paragraph bridging pages 3-4 of the instant specification. For example, FoxM1B is expressed in U2OS osteosarcoma cells, which are of mesoderm origin. See Figure 1C of Wang et al. Mol. Cell. Biol. 25:10875-10894 (2005) (Exhibit B). FoxM1B expression is also detected in insulinoma cells, which were derived from pancreatic neuroendocrine tumors, as well as hepatocellular carcinoma cells, which are of epithelial cells. See Yao et al. J. Biol. Chem. 272:19827-19836 (1997) (Exhibit C). Both references have been cited in the instant application. The neuroendocrine cells and epithelial cells are both derived from endoderm.

8. Voluminous research has confirmed that expression of FoxM1B can be detected at significantly elevated levels in many tumors derived from cells of diverse origins, and that FoxM1B plays a pivotal role in tumor progression. For example, Pilarsky et al., Neoplasia, 6:744-750 (2004) (Exhibit D) demonstrated that FoxM1B is overexpressed in 10 out of 11 different tumors tested, including kidney and breast tumors. See Exhibit D, page 746, right column last paragraph to page 748, left column, first paragraph. Chandran et al., BMC Cancer, 7:64 (2007) (Exhibit E) showed that FoxM1B is expressed in prostate cancer. See Abstract of Exhibit E. Douard et al., Surgery 139:665-670 (2006)

(Exhibit F) showed that FoxM1B is expressed at elevated levels in colonic adenocarcinomas. See Abstract of Exhibit F. Further, Kim et al. described that FoxM1B was expressed in a variety of distinct human cancers including hepatocellular carcinomas, intrahepatic cholangiocarcinomas, basal cell carcinomas, ductal breast carcinomas, anaplastic astrocytomas, glioblastomas, and non-small cell lung cancers. See Abstract of Kim et al., Cancer Res. 66:2153-2161 (2006) (Exhibit G). Glioblastomas and anaplastic astrocytomas were derived from neural cells of ectoderm origin. Thus, FoxM1B is expressed in numerous actively replicating tumor cells of different origins.

9. Additionally, the inhibitory effects of p19ARF 26-44 peptide on FoxM1B activity are seen not only in liver tumor cells, but also in a variety of other FoxM1 B expressing cell lines. For instance, p19ARF protein binds to FoxM1B protein in U2OS osteosarcoma cells as well as in hepatocytes. See Examples 15 and 16 of the instant application. Applicants have also demonstrated that the p19ARF peptide inhibited FoxM1B-depedent tumor progression in osteosarcoma as well as in hepatocarcinoma cells and endothelial cells. See Example 17 of the instant application, Figure 3 of Gusarova et al. J. Clin. Invest. 117:99-111 (2007) (Exhibit H), and Figure 8E of Exhibit H, respectively. Exhibit H has already been submitted to the Office as Exhibit B to the October 26 Declaration. Further, the p19ARF peptide inhibited FoxM1B-dependent transactivation in lung adenocarcinoma A549 cells. See Figure 5B of Wang et al. Oncogene, on-line publication pp. 1-13 (2008) (Exhibit I). Thus, the p19ARF 26-44 peptide inhibits FoxM1B activities in a variety of FoxM1B expressing cells, not limited to liver tumors or tumor cells of epithelial origin.

10. Based on the foregoing, I conclude that FoxM1B is expressed in many proliferating tumor cells of diverse origins, including but not limited to liver tumors and tumor cells of epithelial origin. I believe the evidence shows that FoxM1B plays a pivotal role in tumor cell proliferation. I further conclude that the p19ARF 26-44 peptide inhibits FoxM1B activities in a variety of FoxM1B expressing cells. This evidence, in my opinion, would convince someone knowledgeable in the field of oncology that p19ARF peptide would have broad therapeutic benefits in treating different types of tumors that express FoxM1B at elevated levels.

11. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Respectfully submitted,

Dated: 03/26/2008

By: I-Ching Wang

I-Ching Wang, Ph.D.